Biopharmaceutics of Didanosine in Humans and in a Model for Acid-Labile Drugs, the Pentagastrin-Pretreated Dog

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Didanosine is a purine nucleoside analogue approved for the treatment of human immunodeficiency virus infection. It is extremely unstable at pH values less than 3 and requires protection against gastric acid-induced hydrolysis. Beagle dogs pretreated with pentagastrin, an analogue of gastrin that reproducibly stimulates gastric acid secretion, have been used to screen different didanosine formulations. The absolute bioavailability of didanosine from a saline solution decreased from approximately 43% in untreated dogs to 8% after pretreatment with pentagastrin. Administration of buffered solution of didanosine to untreated and pretreated dogs yielded bioavailability estimates of 37 and 30%, respectively. In humans, the bioavailability from a similar buffered solution was approximately 40%. Pentagastrin-pretreated dogs were used to evaluate four new products relative to a citrate-phosphate buffer sachet, the formulation selected for large-scale clinical trials in humans. Two of these new formulations, a chewable tablet and an antacid suspension, were more bioavailable than the reference sachet. This also proved to be true in man, necessitating an adjustment in the dose of didanosine when administered as the chewable tablet. Dogs pretreated with pentagastrin accurately predicted the improved bioavailability of new didanosine formulations prior to clinical use. This animal model may be helpful in evaluating the biopharmaceutics of other acid-labile drugs.

KEY WORDS: didanosine; pentagastrin-pretreated dog; formulation development; bioavailability.

INTRODUCTION

Didanosine (ddI; Videx) is an antiretroviral agent with activity against the human immunodeficiency virus (HIV). Didanosine requires protection against acid-induced hydrolysis in the stomach through coadministration with appropriate buffering agents (1). When didanosine is administered orally to patients as an aqueous solution with Maalox or mixed with a combination of citrate and phosphate buffers, the absolute bioavailability is approximately 40% (2). The pharmacokinetics of didanosine are linear over the range of 0.8 to 10.2 mg/kg, which includes doses of therapeutic interest (2).

The beagle dog is one of the species studied during the preclinical development of didanosine. After intravenous administration of didanosine over the range of 20 to 100 mg/kg, the pharmacokinetics of didanosine are dose dependent (3). When given orally, the absolute bioavailability of didanosine at a dose of 100 mg/kg is 46% (4). The bioavailability of didanosine from a sodium acetate buffer solution ranges from 43 to 48% over a dose range of 50 to 250 mg/kg (5). The published data (2–4) suggest that the pharmacokinetic profile of didanosine in the dog is similar to that in man in many respects, including rapid absorption and elimination, distribution primarily in total-body water, comparable estimates of absolute bioavailability for a buffered solution, and evidence of active renal tubular secretion in both species.

During the didanosine Phase I and II clinical program, several changes in formulation were made. In order to screen formulations prior to clinical evaluation, the dog was developed as a model. Since the pH of the contents of the dog stomach can range from 0.8 to 8 (6), animals were pretreated with pentagastrin prior to dosing with didanosine. Pentagastrin, an analogue of the natural hormone gastrin, causes a rapid and reproducible decrease in gastric pH (6,7). The present studies describe the use of the pentagastrin-pretreated dog as a model for the evaluation of new didanosine formulations.

MATERIALS AND METHODS

Dosage Forms

Six formulations of didanosine were utilized in studies conducted in beagle dogs or humans. The first formulation given to dogs consisted of a buffered solution containing 0.45 M sodium phosphate dibasic and 0.30 M sodium citrate dihydrate, pH 9.0. Subsequently, a citrate–phosphate buffer sachet formulation was developed for clinical use, which contained, in addition to 250 or 375 mg of didanosine, sodium citrate USP dihydrate, dibasic sodium phosphate anhydrous, citric acid, and 14.5 g sucrose. The total weight of a unit dose was 20 g, and it was reconstituted to form a solution with a pH of approximately 7.5. Later formulations included a chewable tablet, an antacid suspension, an electrolyte solution, and a compression-coated tablet. The chewable tablet formulation contained 125 or 150 mg of didanosine, dihydroxylalumium sodium carbonate, magnesium hydroxide, sodium citrate, and sucrose. Each tablet weighed 3 g. The antacid suspension formulation contained approximately the same ingredients, both qualitatively and quantitatively, as two chewable tablets. Reconstitution of this formulation with water resulted in the formation of a solution of didanosine, although the antacid components remained in suspension. The electrolyte solution formulation, used only in dogs, contained 230 mg didanosine, sodium and potassium bicarbonate, sodium and potassium citrate, and sucrose. The compression-coated tablet product, also administered only to dogs, contained a core of didanosine (approximately 83 mg per tablet) mixed with microcrystalline cellulose, sur-
rounded by sodium bicarbonate, aluminum hydroxide, and magnesium carbonate.

Studies in Beagles

Design of Study 1

A three-way randomized crossover design was employed to evaluate the absolute bioavailability of didanosine from two solutions after oral administration of a 50 mg/kg dose to three adult male beagle dogs. Each dog received an intravenous dose of didanosine over 5 min, administered at the rate of 0.5 mL/min/kg using a calibrated infusion pump. The intravenous solution was prepared in sterile 0.9% sodium chloride and filtered through a 0.22-μm filter. The oral doses of didanosine were administered by gavage as solutions prepared in 0.9% sodium chloride or the pH 9.0 buffer in a volume equivalent to 3.5 mL/kg. After the completion of the initial three treatments, the same dogs received the oral doses after pretreatment with pentagastrin (Peptavlon), delivered intramuscularly at a dose of 6 μg/kg 20 min prior to dosing with didanosine. Dogs were fasted overnight before and for 6 hr after dosing. There was a minimum recovery period of 1 week between sessions.

Dogs were restrained in fabric restraint slings for the first 6 hr after dosing. A 3-mL sample of blood was collected into a heparinized Vacutainer prior to dosing and at 3, 6, 9, 12, 15, 21, 30, and 45 min and 1, 1.5, 2, 3, 4, 6, 8, and 10 hr after the intravenous dose and at 10, 20, 30, and 45 min and 1, 1.25, 1.5, 2, 3, 4, 6, 8, 10, and 12 hr after each oral dose. Blood samples were obtained either from a saphenous vein catheter or via jugular venipuncture. Plasma was prepared within 60 min of sample collection, then stored at −20°C. Catheterized urine samples were obtained predose and over the intervals 0 to 1, 1 to 2, 2 to 3, 3 to 4, and 4 to 6 hr after dosing. After the 6-hr sample, the dogs were transferred to stainless-steel metabolism cages. Two additional urine samples were obtained over the intervals of 6 to 10 and 10 to 24 hr by collecting whatever urine was voided in the cage tray, which drained into a polypropylene container held on ice. The cage was rinsed with approximately 50 mL of water to ensure a complete collection of the voided urine at the end of each interval. After the total volumes of the urine and rinse water were recorded, an aliquot of urine was mixed with 0.02 M potassium phosphate buffer (pH 8.0), 1 part urine to 2 parts buffer. The buffered urine samples were stored at −20°C until analysis.

Design of Study 2

A five-way randomized crossover was conducted in five adult male beagle dogs to evaluate the bioavailabilities of four oral formulations of didanosine, relative to that from the citrate–phosphate buffer sachet used in human studies. Each dog was pretreated with pentagastrin as described in Study 1. A 250-mg dose of each formulation was given after an overnight fast. The four test products administered were the chewable tablet, antacid suspension, electrolyte solution, and compression-coated tablet. Two chewable tablets (125 mg didanosine per tablet) were ground to 20 mesh and mixed with 120 mL of water. The citrate–phosphate buffer, antacid suspension, and electrolyte solution were reconstituted with 120 mL of water prior to administration. Each of these four formulations was administered by gavage, and the gavage tube was rinsed with 20 mL of water prior to removal from the stomach. For the compression-coated tablet, three tablets were given in rapid succession following gavage administration of 140 mL of water. One week elapsed between treatments.

Serial blood samples were collected using jugular venipuncture into heparinized Vacutainers prior to dosing and at 15, 30, and 45 min and 1, 1.5, 2, 3, 4, 6, and 8 hr after dosing. The total urine output from each dog was collected over a 24-hr interval into the cage tray and processed as described in Study 1. Plasma and buffered urine samples were stored at −20°C until analysis.

Studies in Humans

Design of Study I

Based upon their superior bioavailability in the second study conducted in dogs, the chewable tablet and antacid suspension formulations were selected for evaluation in humans. A randomized three-way crossover study, balanced for first order residual effects, was conducted in 18 male patients, using the citrate–phosphate sachet as the reference product. The patients were seropositive for HIV but did not have any symptoms of AIDS or AIDS-related complex. In addition, patients were excluded from the study if there was any evidence of hepatic or renal dysfunction. Patients signed the appropriate informed consent documents prior to any study procedures. The mean (SD) age, body weight, and height of the patients were 31 (3) years, 77.1 (10.1) kg, and 178.6 (6.5) cm, respectively.

Each patient received a single 375-mg dose of each of the three didanosine formulations. Patients were required to fast for 10 hr prior to dosing and for 4 hr after drug administration. There was a 1-week washout period between sessions. The tablets were chewed in rapid succession, followed by a rinse and gargle with 120 mL of room-temperature tap water. The sachet and suspension formulations were reconstituted with 120 mL of water, then swallowed immediately. Serial heparinized blood samples were collected prior to and at 0.15, 0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 5, 6, 8, and 10 hr after dosing. The total urine output, collected as discrete intervals, was obtained predose, 0 to 4, 4 to 8, and 8 to 12 hr after dosing. The collection bottles were kept in a refrigerator except during voiding. At the end of each interval, the total volume of the urine was measured and recorded. The urine sample was mixed thoroughly and a 2-mL portion of the sample was transferred to a polypropylene tube containing 4 mL of 0.20 M potassium phosphate buffer, pH 8.0. Plasma and urine samples were stored at −20°C prior to analysis.

Design of Study II

Since the bioavailability of didanosine from the chewable tablet was greater than that from the buffered sachet, a second bioequivalence study was conducted. In Study II, the dose of the chewable tablet formulation was 20% lower than the dose of the citrate–phosphate buffer sachet. Twenty-four male patients seropositive for HIV were enrolled using a randomized two-way crossover design. The